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Antimicrobial resistance : Patterns and trends in the National University Hospital, Singapore (1989-1991)

G KUMARASINGHE MRCPath, HY LIEW BSc, and C CHOW MSc.

Division of Microbiology, Department of Laboratory Medicine, National University Hospital, Lower Kent Ridge Road, Singapore 0511

Abstract

The commonly isolated organisms, *Staphylococcus aureus, Escherichia coli,* Klebsiella species, *Pseudomonas aeruginosa,* Acinetobacter species, Proteus species and Enterobacter species from clinical material other than blood, cerebrospinal fluid and stool, were analysed for their incidence and increasing trends of resistance to the commonly used antimicrobials.

Staphylococcus aureus was tested against penicillin, methicillin, erythromycin, gentamicin and co-trimoxazole; *Pseudomonas aeruginosa* against amikacin, ceftazidime, gentamicin, piperacillin and cefsulodin; and gram negative bacilli against ampicillin, co-trimoxazole, cephalexin, cefuroxime, ceftriaxone, nalidixic acid and nitrofurantoin.

Methicillin sensitive *Staphylococcus aureus* exhibited a high degree of resistance to penicillin only (83%), but methicillin resistant *Staphylococcus aureus* (34-46%) showed nearly 100% resistance to all drugs tested except for co-trimoxazole over the period of study. A high incidence of resistance was found among Klebsiella species, Enterobacter species, Acinetobacter species and Pseudomonas species. Increasing trends of resistance against cephalosporins were noticed with respect to Acinetobacter species, Klebsiella species and Enterobacter species for cefuroxime and ceftriaxone; *Pseudomonas aeruginosa* for ceftazidime and cefsulodin.

The overall resistance of organisms is notably high. Methicillin-resistant *S. aureus* is endemic and accounts for about 39% of all *S. aureus* isolates. The typical nosocomial organisms like Acinetobacter species and Klebsiella species are increasingly developing resistance to useful drugs such as gentamicin, cephalexin and ceftriaxone.

Key words: Antibiotics, resistance patterns, trends.

INTRODUCTION

Studies of bacteria from "antibiotic virgin" populations or bacteria isolated from the pre-antibiotic era (common pathogens such as *Staphylococcus aureus, Escherichia coli,* and other bacteria) generally show a low or absent prevalence of resistance.[1] Emergence of resistance of the infecting organism during antibiotic therapy is a well known phenomenon which exists since antibiotics were introduced on the market.[2] Ever since the introduction of the early forms of sulphonamides and penicillin some microbes have proved resistant to one or more drugs.[3] The previously susceptible organisms have become resistant; some by means of well-understood mechanisms. others by ways yet to be described.

In serious infections antimicrobial therapy should be started before culture results are available. Patterns of antibiotic use and bacterial resistance vary from country to country, and even between neighbouring hospitals? Clearly, therefore, rational guidelines for prescribing will hinge on up-to-date information about local prescribing habits and the prevalence and sensitivity of the local bacteria.

The purpose of this project was two-fold. Its first objective was to establish the prevalence of species that are pathogenic to patients in the hospital and its second aim was to demonstrate the extent of resistance to antimicrobials.

MATERIALS AND METHODS

The National University Hospital is a teaching hospital admitting patients from inland and overseas, to the usual range of specialities. It opened in mid-1985, and grew gradually to reach peak utilization of its 700 beds by the beginning of 1989.

Most frequently isolated clinically signifi-

Address for correspondence and reprint requests: G. Kumarsinghe, Division of Microbiology. Department of Laboratory Medicine, National University Hospital, Lower Kent Ridge Road, Singapore 0511

cant bacteria from patients' samples from sites other than blood, CSF and stool during the period January 1989 to December 1991 were included in this study. The organisms isolated from blood cultures were analysed for a variety of antibiotics in a separate study conducted in the hospital. Both in-patients and out-patients who attended the hospital were considered; about 20% of specimens were derived from out-patients. Specimens from patients were collected for clinical indications, as determined by the attending clinician.

Bacteriological cultures were carried out by plating on the following media.

Urine	:	CLED agar
Respiratory	:	Horse blood agar, chocolate agar
Pus (all other sites except CSF, blood and stool)	:	Horse blood agar, MacConkey agar

The cultures were incubated under aerobic and anaerobic conditions at 37°C overnight. Identifications were made by conventional biochemicals, commercial kits (API and Microbact) and serodiagnostic tests.

Antimicrobial susceptibility testings were performed for the commonly used drugs (first line) on Mueller-Hinton agar (Becton Dickinson) by the disk diffusion method of Bauer *et al.*[5] *Staphylococcus aureus* was tested for methicillin resistance using a 5mcg disc on Mueller Hinton agar incorporated with 5% of sodium chloride and incubated at 35-37°C for 18 hours.

The methods were controlled by internal and external quality assessment. The laboratory participates in quality control programmes organised by the U.K. National External Quality Assessment Scheme for Microbiology and the College of American Pathologists.

RESULTS

Prevalence of pathogenic species isolated

The project was initiated and completed between January 1989 and December 1991. A total of 18,560 pathogenic bacteria isolated from clinical material (other than CSF, blood and stool) derived from patients in the National University Hospital were included in the study. Wherever possible, repeat culture results were excluded to avoid potential bias.

Stuphylococcus aureus formed the most frequently recovered organism at an incidence rate of 19.8%, followed closely by *Escherichia coli* and Klebsiella species at 19.5% and 16.0% respectively. On the whole, gram-negative organisms accounted for two-thirds of the total population isolated (Table 1).

Over the three years, isolation of *Staphylococcus aureus* showed a significant decrease ($p < 0.001$; chi-square test for trend) while *Streptococcus Group* D showed a steady increase. The rest of the organisms did not show any significant change (Fig. 1).

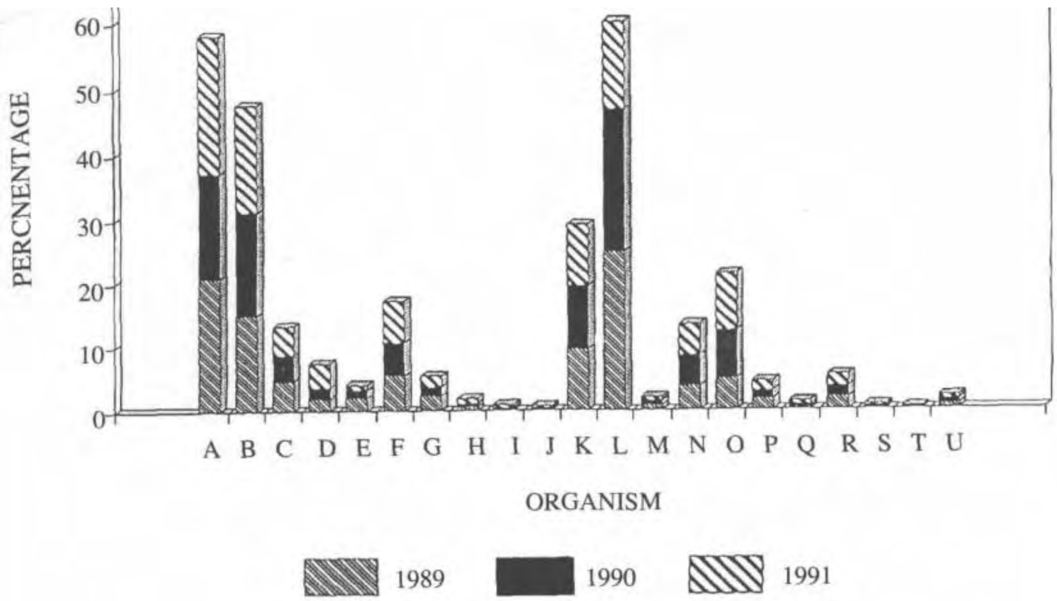
From the observations made in Table 1, the top six gram- negative rods, namely *Escherichia coli*, Klebsiella species, *Pseudomonas aeruginosa,* Acinetobacter species, Proteus species and Enterobacter species and the most prevalent *Staphylococcus aureus*, were identified for further establishment of the extent of their resistance to the commonly used antimicrobial~.

All Pseudomonas species other than aeruginosa and pseudomallei were analysed as a group. Although these species were low in prevalence, their reputation as nosocomial pathogens, especially to immunocompromised patients, justified their inclusion to the list of organisms above.

TABLE 1: Incidence of microbial flora isolated from body sites other than blood, CSF and stool from NUH, 1989-1991

Organism	Incidence (%)
Staphylococcus aureus	19.8
Escherichia coli	19.5
Klebsiella species	16.0
Pseudomonas aeruginosa	9.8
Streptococcus group D	7.2
Acinetobacter species	5.8
Proteus species	4.5
Streptococcus group B	4.5
Enterobacter species	2.6
Haemophilus influenzae	1.9
Citrobacter species	1.9
Streptococcus, other gps	1.5
Pseudomonas species	1.3
Streptococcus group A	0.7
Others	0.7
Morgenalla morganii	0.6
Streptococcus pneumoniae	0.4
Serratia species	0.4
Moraxella catarrhalis	0.4
Aeromonas species	0.3
Neisseria gonorrhoeae	0.2

Total number of isolates = 18,560



A. ***Escherichia coli***
B. Klebsiella species
C. Proteus species
D. Enterobacter species
E. Pseudomonas species
F. Acinetobacter species
G. Citrobacter species
H. ***Morganella morganii***
I. Serratia species
J. Aeromonas species
K. ***Pseudomonas aeruginosa***
L. *Staphylococcus aureus*
M. Streptococcus group A
N. Streptococcus group B
O. Streptococcus group D
P. Beta-streptococcus, other
Q. ***Streptococcus pneumoniae***
R. ***Haemophilus influenzae***
S. ***Moraxella catarrhalis***
T. ***Neisseria gonorrhoeae***
U. Others

FIG. 1: The trend of isolation incidence of clinically significant isolates from sites other than blood, CSF and stool, from 1989-1991.

Streptococcus group D and *Streptococcus group* B were the fifth and eighth most isolated organisms respectively. In a separate study of bacteraemia causing organisms done in the hospital, *Enterobacteriaceae* and staphylococci constituted the largest groups out of some 40 different types of organisms.

Extent of resistance of commonly isolated organisms to the commonly used antimicrobials

Escherichia coli and Proteus species were most susceptible to gentamicin, cefuroxime and ceftriaxone while their resistances to ampicillin, co-trimoxazole and cephalexin have surpassed the 30% mark.

The resistance patterns of Klebsiella species and Enterobacter species were even more alarming. They exceeded 30% in resistance to all the commonly used antimicrobials under study.

By and large, most Enterobacteriaceae were encountering increasing trends of resistance to the cephalosporins (Figs. 2A-D).

Acinetobacter species showed a high degree of multiple resistance that exceeded 50% for all the six antimicrobials tested. There was also a significant increase in its resistance towards the cephalosporins.

Except for co-trimoxazole, Pseudomonas species were found to be exhibiting resistances that were either approaching 50% or beyond (Figs. 2E-F).

Methicillin-resistant *Staphylococcus aureus* (MRSA) accounted for about 40% of the *Staphylococcus aureus* population in the hospital and significant increase in resistance (30%-55%) was observed to co-trimoxazole among them (Fig. 3A). Erythromycin and gentamicin resistance remained over 98%. Among the methicillin-sensitive group there was no problem of resistance except with penicillin (81%-85%).

A steady increase in resistance to ceftazidime and cefsulodin by *Pseudomonas aeruginosa* was noted while aniikacin remained the most susceptible antimicrobial (Fig. 3B).

Most Enterobacteriaceae, especially *Escherichia coli* and Klebsiella species were the most likely causative agents of uncomplicated lower urinary tract infections. With ampicillin resistance exceeding 50% and co-trimoxazole still increasing beyond 35%, their places as the first choice of drugs may need to be reassessed in comparison with other alternatives such as nalidixic acid and nitrofurantoin (Fig. 4).

Most of the commonly isolated gram-negative rods exhibited an increasing trend in resistance to the cephalosporins. *Escherichia coli* and Klebsiella species showed a marked increase in resistance to cephalexin from 1989 to 1990 while Klebsiella species, Enterobacter species and Acinetobacter species indicated steady increases in resistance to cefuroxime and ceftriaxone.

Gentamicin remained an effective drug against Proteus species and *Escherichia coli*. However, the resistance pattern of the rest of gram-negative rods remained high at 30% or above, during the period of study.

13-15% of *Streptococcus group* D and 18-44% of *Haemophilus influenzae* were resistant to ampicillin.

The sensitivity of organisms isolated from blood cultures followed a pattern similar to those isolated from other sites.

DISCUSSION

The most commonly isolated organisms were *Staphylococcus aureus*, *Escherichia coli*, Klebsiella species and *Pseudomonas aeruginosa*, with *Streptococcus group* D and Acinetobacter species showing steady increases annually. The significant decrease in *Staphylococcus aureus* isolation could be attributed to a Hospital Infection Control hand-washing drive that was conducted sometime in mid-1990. During the period of study we have observed similar increases in the incidence of *Streptococcus group D*[6] and Acinetobacter species[7] infections as seen in other centres. Over the years these organisms have become important causes of infection because of the increasing frequency and severity of associated infections in intensive care units, and because of their antibiotic resistance.

The newer drugs are rarely more efficacious against susceptible organisms than the older preparations. We wish to highlight the position regarding the use of common antibiotics, often reported as "First Line" antimicrobials by the microbiologists. Hospital policy encourages the use of antimicrobials that are less toxic and less expensive and restricts the use of newer cephalosporins and quinolones to clear indications. Clearly there has been a rapid change over the period, which also saw the increasing use of third-generation cephalosporins in hospital practice (Fig 5).

Some of the frequently used antimicrobials, such as ampicillin and cephalexin are encountering resistance levels in excess of 50% to all

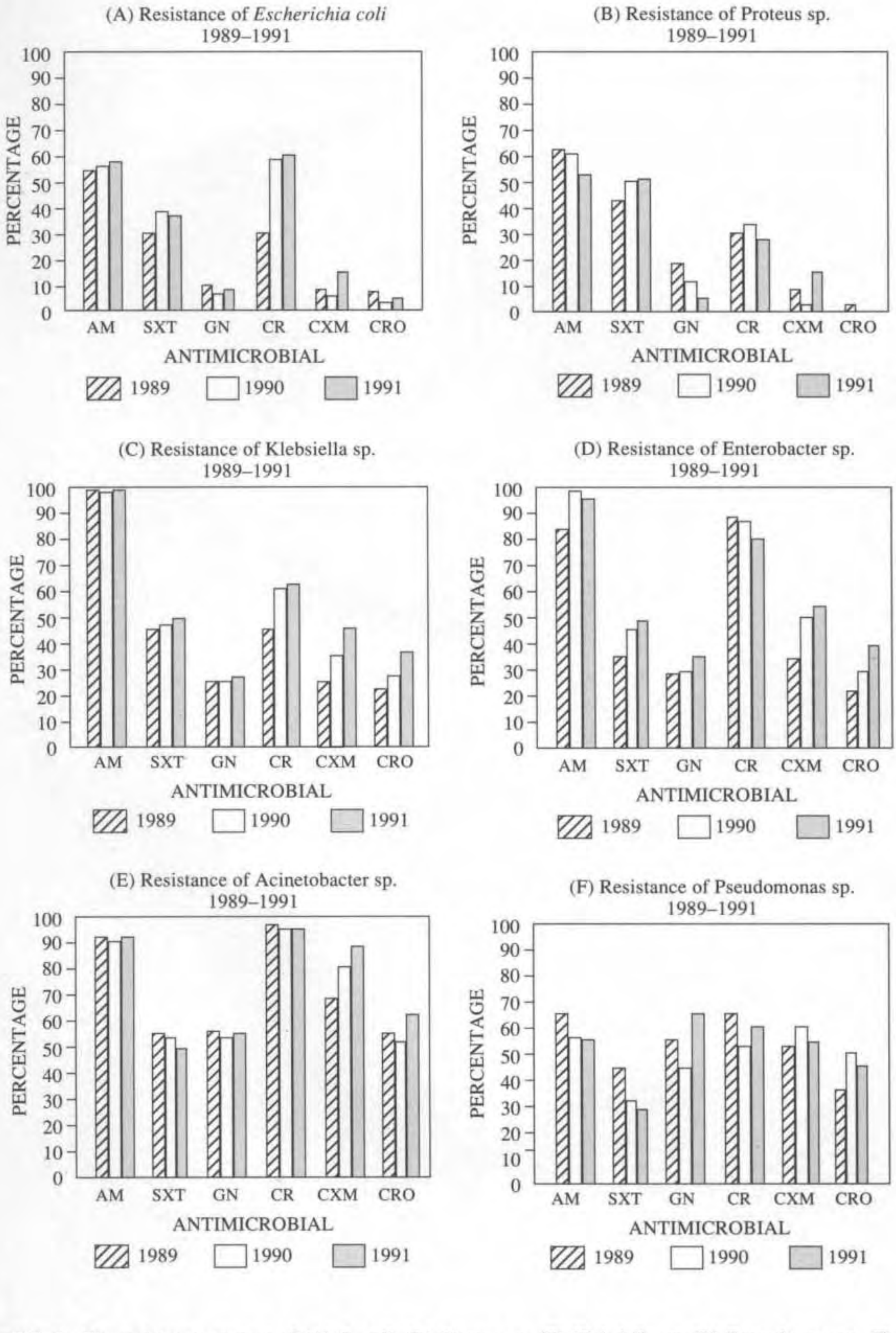


FIG. 2: The resistance patterns of (A) E. coli, (B) Proteus sp, (C) Klebsiella sp, (D) Enterobacter sp, (E) Acinetobacter sp, (F) Pseudomonas sp, against AM = ampicillin, SXT = co-trimoxazole, GN = gentamicin, CR = cephalexin, CXM = cefuroxime, CRO = ceftriaxone.

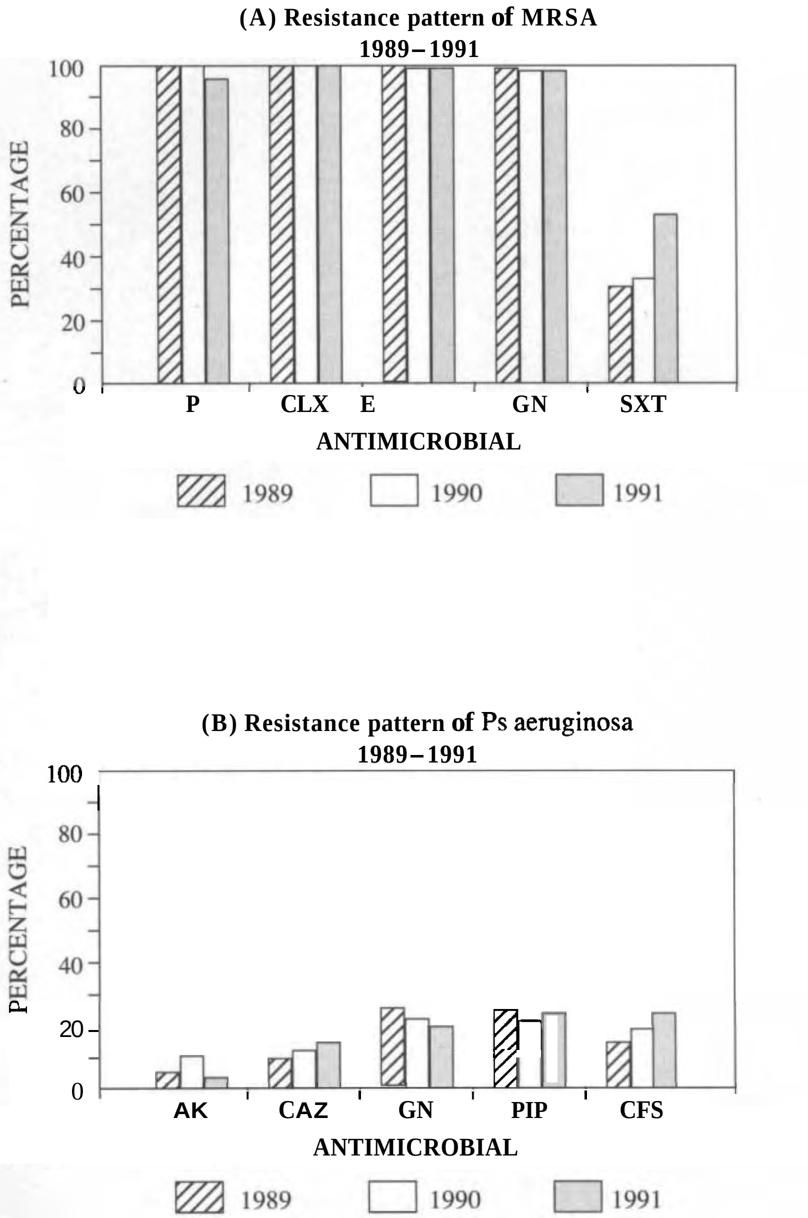
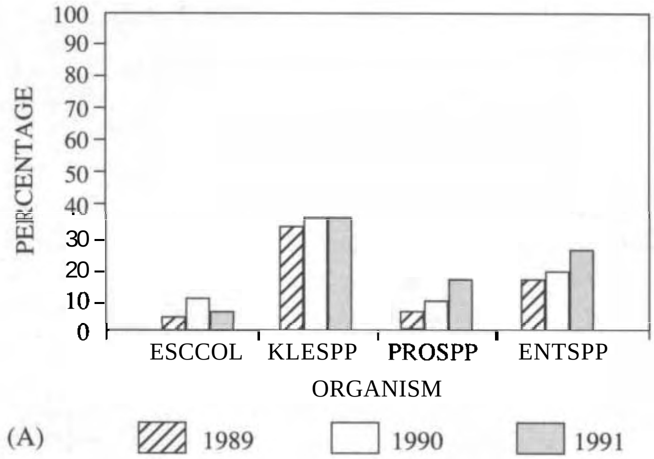


FIG. 3: The resistance patterns of (A) methicillin-resistant *Staphyloccocus aureus* against P = penicillin, CLX = cloxacillin, E = erythromycin, GN = gentamicin, SXT = co-trimoxazole; (B) *Pseudomonas aeruginosa* against AK = amikacin, CAZ = ceftazidime, GN = gentamicin, PIP = piperacillin and CFS = cefsulodin.



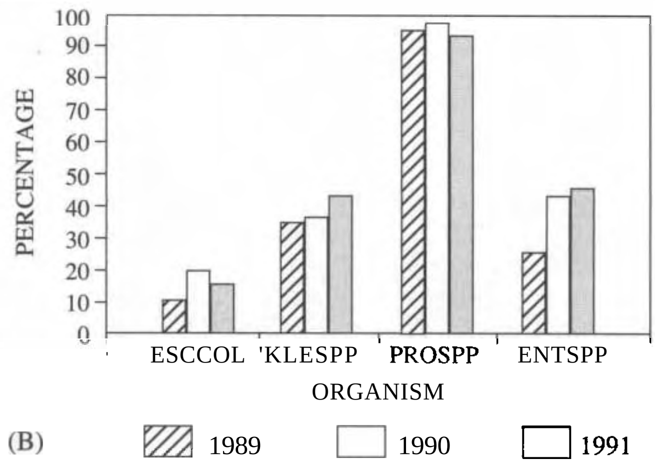


FIG. 4: Resistance patterns of urinary gram-negative bacilli ESCCOL = ***Escherichia coli***, KLESPP = Klebsiella sp, PROSPP = Proteus sp, ENTSPP = Enterobacter sp against (A) nalidixic acid; (B) nitrofurantoin.

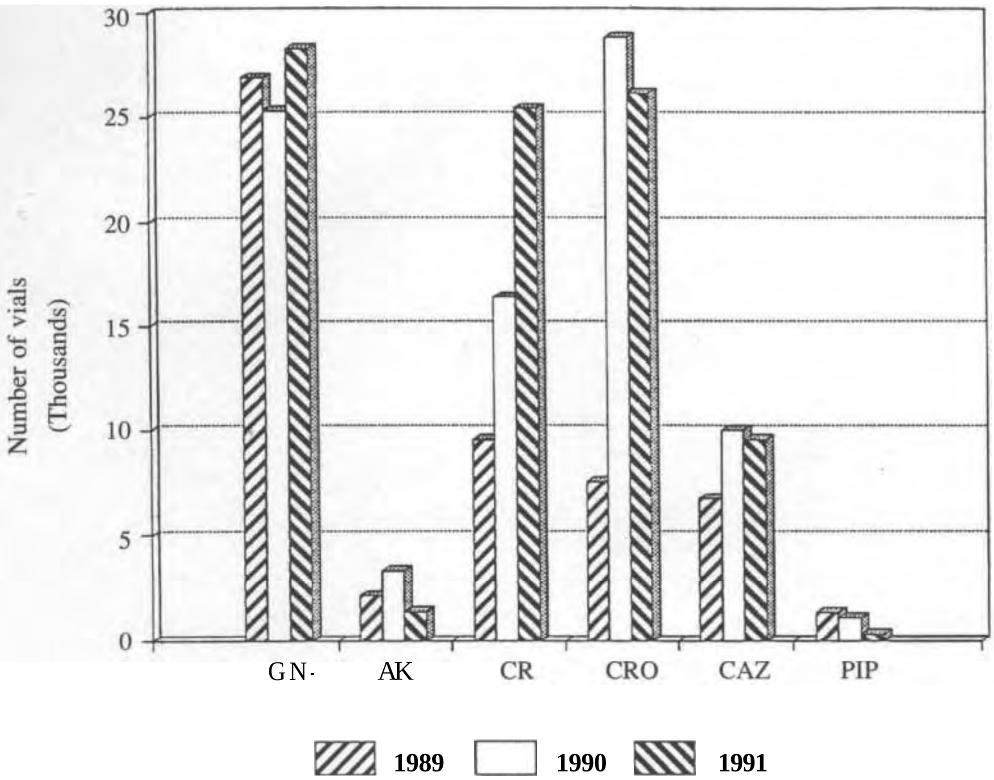


FIG. 5: Antibiotic usage (injectable preparation) in the hospital from **1989-1991.** GN = gentamicin, AK = amikacin, CR = cefalexin, CRO = ceftriaxone, CAZ = ceftazidime, PIP = piperacillin.

the pathogens under study except Proteus species, and gentamicin and ceftriaxone are already encountering resistance levels approaching 40% with Klebsiella species and Enterobacter species and much higher levels against Acinetobacter species and Pseudomonas species. Methicillin-resistant ***Staphylococcus aureus*** has become an intractable problem in the National University Hospital as well, as in many other hospitals in the world.[8] To the extent that community-acquired organisms will have been included in Figs. 2-4, these figures will understate the increasing prevalence of resistance among hospital acquired strains.

Acinetobacter species survive very well in a dry or moist environment and even in the air. Survival of these organisms on common objects, such as patients' charts may be relevant to the spread of infection.[7] The drug resistance is alarming with more than 50% resistance to all the drugs tested. If the increasing trend continues, it may become a major pathogen in the hospital, causing treatment problems.

Different reasons may be offered for the increased resistance rates observed in the present study. The significant increase in ampicillin resistance probably reflects the widespread use of this in the past and increased rates of gentamicin and cephalosporin resistance with Acinetobacter species, Klebsiella species and Enterobacter species may reflect the emergence of plasmid mediated resistance.

There has been an alarming increase in the usage of cephalosporins in the National University Hospital over the three years of study (Fig. 5). This has made a significant impact to the increasing resistances to most of the gram-negative organisms under study, and also probably aided the emergence of MRSA.[9]

Useful drugs such as nitrofurantoin and nalidixic acid are being used sparingly in the treatment of urinary tract infections although their costs and resistance patterns are comparatively low.

This study has demonstrated that resistance to antimicrobial agents must be taken into consideration when treating respiratory, wound and urinary infections associated with the presence of these species.

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